June 20, 1949.

Dear Max:

Your isolates of H168 were received in very good shape, and I had no trouble reisolating the heterozygote. They all agree in being pure Xyl-, although Lacy and Mtly. The trouble I reported having with reisolating the heterozygotes from the cultures you sent stems, I think, from my presumption that they were Xyly, which they apparently have not been since they reached you.

"8"-217 is being checked for its nutrition now; it is certainly a Lac#Mtl- segregant. Are you surprised that it should have a lethal sib?, or just that it was not predicted as such?

I really don't know what to make of the H-168 Xylose story. I have an old slant of it, which does contain Xyl# cells, but I haven't been able to reisolate any heterozygotes from it, either on lactose or mannitol. This tube was a duplicate of the one I sent you, and I am a little hesitant about accepting the notion that this is all a clerical error. I had just burnt my fingers then, you may remember, about H-72! However, unless you happen to have earlier cultures from H-168, preferably the one I sent you, to check up on the presence of Xyl# cells, I don't know how it could be decided. Have you ever noted the presence of Xyl# cells in any derivative of 168??

Change of type could be a trivial coincidence. Segregation results in large numbers of cells with diverse requirements, and the technique used in reisolating y stocks is identical with that used in the original isolation. Conceivably, the new H168 is the result of a new crossing. I am trying to reproduce the whole phenomenon with some new isolates which are very similar to the original 168.

This week, I've finally isolated not one, but <u>two</u> stocks which are heterozygous for Mal_1; at the same time have repeated, affirmatively, reversion experiments showing that this same Mal_1 is usually hemizygous. Although Mal_1 y, is very rare, they do occur sometimes, and make the whole story that much harder to interpret. One of the Mal y came out of a cross (Lac_1- x Lac_4-) like that described in the PNAS paper, not involving <u>Het</u>. Since this stock is heterozygous for Lac, Mal, Xyl, Mtl, and Gal, and is a "spontaneous" heterozygote, it is certainly the culture of choice for the study of the latter. I'll do some work on the segregation pattern while you're on your trips, and send <u>H-215</u> on to you later.

Don't distract yourself from your vacation, but if you can remember anything else about H-168, I'd appreciate hearing it.

Sincerely,

Joshua